

Carbon-13 Fractionation of Relic Soil Organic Carbon during Mineralization Effects Calculated Half-Lives

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The ^{13}C natural abundance approach for determining soil organic C (SOC) stability and turnover has been used to determine SOC mineralization kinetics. These calculations generally assume that ^{13}C fractionation during relic SOC and unharvested biomass mineralization is insignificant. The objective of this study was to determine the impact of this assumption on calculated relic SOC half-lives. Study sites were located in Minnesota and South Dakota. At the Minnesota site, SOC contained in the surface 30 cm of soil in a fallowed area decreased from 90.8 to 73.2 Mg ha^{-1} during a 22-yr period. Associated with this decrease was a 0.72‰ increase in the soil $\delta^{13}\text{C}$ values (from -18.97 to -18.25 ‰). Based on these values, the Rayleigh fractionation constant (ϵ) of relic SOC was -3.45 ‰. At the South Dakota site, SOC decreased 10% ($2.8 \pm 1.8 \text{ g kg}^{-1}$) and $\delta^{13}\text{C}$ increased 3.2‰ (0.548 ± 0.332 ‰) during a 5-yr period. The Rayleigh fractionation constant for this experiment was -6.94 ‰ (± 4.74 ‰). In a separate experiment, the $\delta^{13}\text{C}$ value of corn (*Zea mays* L.) and soybean [*Glycine max* (L.) Merr.] residue remained unchanged after 4 mo. The impact of ^{13}C enrichment during relic C mineralization on calculated C budgets depends on the type of residue returned to the soil. A simulation study showed that for systems where C_4 residues are returned to soil derived from C_3 and C_4 plants, not considering ^{13}C enrichment during relic SOC mineralization will result in underestimating relic SOC half-lives and overestimating the contribution of fresh C_4 biomass in the SOC. The effect of ^{13}C enrichment during relic SOC and unharvested biomass mineralization had cumulative impacts on C budgets and did not cancel each other out. The reverse was true for C_3 biomass. To minimize these errors, SOC maintenance rate experiments should measure ^{13}C enrichment during relic SOC and unharvested biomass mineralization.

Abbreviations: NHC_a , the amount of non-harvested C applied; PCR, plant biomass C returned to soil; $\text{PCR}_{\text{incorp}}$, new biomass C incorporated into SOC; SOC, soil organic carbon; $\text{SOC}_{\text{retained}}$, the amount of soil organic carbon retained in soil after mineralization; $\text{SOC}_{\text{final}}$, soil organic carbon contained in soil at the end of the experiment; $\text{SOC}_{\text{initial}}$, soil organic carbon at the beginning of the experiment; SOC_{lost} , the amount of organic C lost; $\delta^{13}\text{C}_{\text{soil final}}$, $\delta^{13}\text{C}$ value of soil at the end of the experiment; $\delta^{13}\text{C}_{\text{PCR}}$, $\delta^{13}\text{C}$ value of plant material remaining in soil after mineralization; $\delta^{13}\text{C}_{\text{SOC retained}}$, $\delta^{13}\text{C}$ of soil organic carbon at the beginning of the experiment that is retained in the soil after mineralization; ϵ , Rayleigh fractionation coefficient.

To minimize erosion and improve long-term sustainability, SOC must be maintained. Soil organic C maintenance experiments typically rely on measuring nonisotopic changes in SOC with time and using differences in ^{13}C isotopic discrimination in C_3 and C_4 plants to quantify C transfers across trophic levels. The nonisotopic approach to estimate SOC maintenance requirements is based on the equation

$$\frac{\text{NHC}_a}{\text{SOC}_e} = \frac{(1/k_{\text{NHC}})\text{SOC}_e}{d\text{SOC}/dt} + \frac{k_{\text{SOC}}}{k_{\text{NHC}}} \quad [1]$$

where NHC_a is unharvested biomass C returned to the soil, SOC_e is soil organic C at the equilibrium point, k_{SOC} is the mineralization rate of soil organic C, and k_{NHC} is the mineralization rate of unharvested C (Clay et al., 2006). Maintenance rates using Eq. [1] are calculated by: (i) defining $\text{NHC}/(\text{SOC at the beginning of the experiment})$ as y and $(d\text{SOC}/dt)$ as x ; (ii) fitting the data to a zero-order rate equation; and (iii) multiplying SOC times the y intercept. Sensitivity analysis of the nonisotopic approach showed that to accurately determine C budgets and maintenance rates, accurate estimates of below-ground biomass were needed (Clay et al., 2006).

The ^{13}C natural abundance method for estimating C budgets has been used in systems where plants growing in the soil have different $\delta^{13}\text{C}$ values than the soil. This approach is based on several premises. First, plants with different photosynthetic pathways have different $^{13}\text{C}/^{12}\text{C}$ isotopic ratios. Second, biological discrimination during the mineralization process is insignificant when compared with the amount of

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discrimination that occurs during photosynthesis (Balesdent et al., 1988). This study investigated the ramifications associated with the second assumption. The objectives of this study were to: (i) quantify the amount of ^{13}C isotopic discrimination that occurs during relic SOC and fresh biomass mineralization; (ii) propose a technique for considering ^{13}C isotopic discrimination during relic and fresh biomass mineralization on C budgets and calculated half-lives; and (iii) determine the potential impacts of ^{13}C fractionation during relic SOC and unharvested biomass mineralization on calculated soil C budgets and SOC half-lives.

MATERIALS AND METHODS

Field Experiment

Carbon-13 Isotopic Fractionation during Mineralization

Research sites were located in Minnesota and South Dakota. These sites were routinely cultivated to prevent plant growth. The Minnesota data were previously reported in Clapp et al. (2000) and Dolan et al. (2006). The soil was a Waukegan silt loam (fine-silty over sandy or sandy-skeletal, mixed, superactive, mesic Typic Hapludoll), and the parent materials were a silt loam loess cap (50–80 cm thick) over neutral to calcareous glacial outwash sand and gravel. Initial soil samples (0–15- and 15–30-cm depths) were collected in 1980 (Clay et al., 1989). During the 22 yr of the study, clean tillage was used to prevent plant growth in the alleyways that were approximately 6 m wide. The north and south end of the alleyways were sampled in 1993 (Clapp et al., 2000) and 2002 (Dolan et al., 2006). Bulk densities were determined as reported by Clapp et al. (2000) and Dolan et al. (2006) when soil samples were collected. Composite soil samples were passed through a 2-mm sieve, stones were removed, and roots and residue returned to the soil samples. Ground samples were ball-milled and duplicate samples analyzed on an elemental analyzer (Carlo Erba, Model NA 1500, Milan, Italy) and stable isotope ratio mass spectrometer (Fisons Optima Model, Fisons Middlewich, UK). The $\delta^{13}\text{C}$ values of soil samples were determined using the equation

$$\delta^{13}\text{C} = 1000 \frac{R_{\text{sample}} - R_{\text{reference}}}{R_{\text{reference}}} \quad [2]$$

where R is the ratio of the heavy to light isotopes in the sample and standard (PDB, $R = 0.0112372$) (Clapp et al., 2000). The working standards were urea ($\delta^{13}\text{C}$ value of -18.2‰) and soil (-17.6‰). The CV for the $\delta^{13}\text{C}$ values was 2.8%. Soil organic C on a volume basis was calculated with the measured mass fraction of C and measured bulk density.

Soil at the South Dakota site was a Divide loam (fine-loamy over sandy or sandy-skeletal, mixed, superactive, mesic, Pachic Calciaquoll). The site is located at $44^{\circ}21' \text{ N}$, $96^{\circ}49' \text{ W}$. The soil pH (1:1 water) was ~ 7.5 . Soil samples from the 0- to 7.5- and 7.5- to 15-m depths were collected from six different sites in 2000 and 2005. Each sample consisted of 10 individual cores. Samples did not visibly react with 0.5 M HCl. Samples were dried, ground (2 mm), and analyzed for total C and $\delta^{13}\text{C}$ on a Europa 20–20 ratio mass spectrometer (SerCon, Crewe, UK). Each sample was analyzed in duplicate. The working standard was wheat (*Triticum aestivum* L.) flower, which had a $\delta^{13}\text{C}$ of -24.64‰ . The standard deviation of the working standards were generally $<1\text{‰}$. Means and 90% confidence intervals for samples collected in 2000 and 2005 were determined. To determine changes in total C and $\delta^{13}\text{C}$ during the 5-yr period, a paired t -test ($P = 0.05$) was used. In this analysis, the values of the samples collected from the same area at the two dates were subtracted from each

other and the mean difference was tested to determine if it was different from zero. The 95% confidence intervals for individual means were determined.

To assess ^{13}C discrimination during corn and soybean residue mineralization, 50 g of dry material, contained in residue bags, was placed on the soil surface. Ten bags of each residue type were used in the study. The $\delta^{13}\text{C}$ value of the initial corn and soybean residue was -11.80 and -27.20‰ , respectively. The C/N ratio for the initial corn and soybean residue was approximately 42:1 and 48:1, respectively. Bags were placed in the field on 17 June 2005 and removed on 25 Oct. 2005. After removing any soil sticking to the residue bag surface, the bags were dried and weighed, gently rinsed with water over a 0.152-mm (100 mesh) screen, dried, ground, and analyzed for $\delta^{13}\text{C}$, total C, and total N. Means and 95% confidence intervals for total biomass, total C, C/N ratio, and $\delta^{13}\text{C}$ were determined. A paired t -test ($P = 0.05$) was used to assess changes in $\delta^{13}\text{C}$ with time.

Determining Carbon Budgets

The relic C half-life calculations were based on the following mass balance equations:

$$\delta^{13}\text{C}_{\text{soil final}} = \frac{\text{PCR}_{\text{incorp}} (\delta^{13}\text{C}_{\text{PCR}}) + \text{SOC}_{\text{retained}} (\delta^{13}\text{C}_{\text{SOC retained}})}{\text{PCR}_{\text{incorp}} + \text{SOC}_{\text{retained}}} \quad [3]$$

$$\text{SOC}_{\text{final}} = \text{PCR}_{\text{incorp}} + \text{SOC}_{\text{retained}} \quad [4]$$

$$\text{SOC}_{\text{initial}} = \text{SOC}_{\text{retained}} + \text{SOC}_{\text{lost}} \quad [5]$$

where $\text{SOC}_{\text{initial}}$ is the SOC in the soil at the beginning of the experiment, SOC_{lost} is the amount of SOC mineralized, $\text{SOC}_{\text{final}}$ is SOC at the end of the study, $\delta^{13}\text{C}_{\text{soil final}}$ is the $\delta^{13}\text{C}$ value of SOC when the experiment was completed, $\text{PCR}_{\text{incorp}}$ is the plant C retained in the soil that was incorporated into SOC, $\delta^{13}\text{C}_{\text{PCR}}$ is the $\delta^{13}\text{C}$ value of the plant material retained in the soil after mineralization, $\text{SOC}_{\text{retained}}$ is the amount of relic C ($\text{SOC}_{\text{initial}}$) retained in the soil at the end of the study, and $\delta^{13}\text{C}_{\text{SOC retained}}$ is the associated $\delta^{13}\text{C}$ value. By simultaneously solving Eq. [3] and [4], the equations

$$\text{SOC}_{\text{retained}} = \frac{\text{SOC}_{\text{final}} (\delta^{13}\text{C}_{\text{soil final}} - \delta^{13}\text{C}_{\text{PCR}})}{\delta^{13}\text{C}_{\text{SOC retained}} - \delta^{13}\text{C}_{\text{PCR}}} \quad [6]$$

$$\text{PCR}_{\text{incorp}} = \frac{\text{SOC}_{\text{final}} (\delta^{13}\text{C}_{\text{soil final}} - \delta^{13}\text{C}_{\text{SOC retained}})}{\delta^{13}\text{C}_{\text{PCR}} - \delta^{13}\text{C}_{\text{SOC retained}}} \quad [7]$$

were derived. If it is assumed that ^{13}C fractionation during SOC and PCR mineralization is minimal, i.e., $\delta^{13}\text{C}_{\text{SOC retained}} = \delta^{13}\text{C}_{\text{soil initial}}$ and $\delta^{13}\text{C}_{\text{PCR}} = \delta^{13}\text{C}_{\text{plant}}$, then Eq. [7] can be simplified into the expression

$$\text{PCR}_{\text{incorp}} = \frac{\text{SOC}_{\text{final}} (\delta^{13}\text{C}_{\text{soil final}} - \delta^{13}\text{C}_{\text{soil initial}})}{\delta^{13}\text{C}_{\text{plant}} - \delta^{13}\text{C}_{\text{soil initial}}} \quad [8]$$

This equation can be solved if soil and plant material collected at time zero ($\delta^{13}\text{C}_{\text{soil initial}}$ and $\delta^{13}\text{C}_{\text{plant}}$) and soil collected at the end of the

experiment are analyzed for total C and $\delta^{13}\text{C}$ ($\text{SOC}_{\text{final}}$ and $\delta^{13}\text{C}_{\text{soil final}}$). Equation [8] can be reorganized into

$$\frac{\text{PCR}_{\text{incorp}}}{\text{SOC}_{\text{final}}} = \frac{\delta^{13}\text{C}_{\text{soil final}} - \delta^{13}\text{C}_{\text{soil initial}}}{\delta^{13}\text{C}_{\text{plant}} - \delta^{13}\text{C}_{\text{soil initial}}} \quad [9]$$

where the ratio between $\text{PCR}_{\text{incorp}}$ and $\text{SOC}_{\text{final}}$ was the relative proportion (p) of new C incorporated in SOC ($p = \text{PCR}_{\text{incorp}}/\text{SOC}_{\text{final}}$). By replacing $\delta^{13}\text{C}_{\text{soil initial}}$ with δ_{C3} , $\delta^{13}\text{C}_{\text{plant}}$ with δ_{C4} , and $\delta^{13}\text{C}_{\text{soil final}}$ with δ , the equations

$$p = \frac{\delta - \delta_{\text{C3}}}{\delta_{\text{C4}} - \delta_{\text{C3}}} \quad [10]$$

$$\delta = p\delta_{\text{C4}} - (1 - p)\delta_{\text{C3}} \quad [11]$$

reported in Wolf et al. (1994) were derived. This derivation shows that Eq. [8, 9, 10, 11] are based on the assumption that ^{13}C discrimination during SOC and unharvested biomass mineralization is minimal. Equations [8, 9, 10, 11] have been used in numerous studies (Balesdent et al., 1988; Follett et al., 1997; Huggins et al., 1998; Collins et al., 1999; Clapp et al., 2000; Allmaras et al., 2004; Clay et al., 2005; Zach et al., 2006). Equation [6] contains three values ($\text{SOC}_{\text{retained}}$, $\delta^{13}\text{C}_{\text{PCR}}$, and $\delta^{13}\text{C}_{\text{SOC retained}}$) that are unknown and therefore to derive a solution for Eq. [6], three independent equations must be solved simultaneously. The first two are Eq. [5] and [6]. The third is the Rayleigh equation:

$$\delta^{13}\text{C}_{\text{SOC retained}} = \delta^{13}\text{C}_{\text{soil initial}} + \epsilon_{\text{SOC}} \ln(\text{SOC}_{\text{retained}}/\text{SOC}_{\text{initial}}) \quad [12]$$

where ϵ is the Rayleigh fractionation constant (Balesdent and Mariotti, 1996). This equation can also be used to calculate the $\delta^{13}\text{C}$ value of the unharvested biomass after mineralization ($\delta^{13}\text{C}_{\text{PCR}}$). The Rayleigh equation has been used to explain isotopic fractionation in a variety of biological systems (Balesdent and Mariotti, 1996; Accoe et al., 2002; Fukada et al., 2003; Spence et al., 2005).

To solve Eq. [5, 6, 12], an iterative approach was used. After stability in the individual C pool sizes, first-order mineralization rate constant (k), half-lives of SOC, and residence times were determined using

$$k = -\frac{\ln(\text{SOC}_{\text{remaining}}/\text{SOC}_{\text{initial}})}{\text{number of years}} \quad [13]$$

$$t_{\text{half-life}} = \frac{\ln(0.5)}{k} \quad [14]$$

$$\text{residence time} = 1/k \quad [15]$$

Potential Impacts of Carbon-13 Discrimination on Half-Life Calculations

Three different systems were used in this analysis. For Model System 1, corn (a C_4 plant) was grown in soil derived from C_3 and C_4 plants. For this system, the values for $\text{SOC}_{\text{soil initial}}$, $\text{SOC}_{\text{soil final}}$, $\delta^{13}\text{C}_{\text{soil initial}}$, $\delta^{13}\text{C}_{\text{soil final}}$, and $\delta^{13}\text{C}_{\text{plant}}$ were 96.25 Mg ha^{-1} , 91.4 Mg ha^{-1} , -19.06‰ , -18.754‰ , and -12.0‰ , respectively. For Model System 2, soybean (a C_3 plant) plants were sown into soil derived from C_3 and C_4 plants. In this system, the values for $\text{SOC}_{\text{soil initial}}$, $\text{SOC}_{\text{soil final}}$, $\delta^{13}\text{C}_{\text{soil initial}}$, $\delta^{13}\text{C}_{\text{soil final}}$,

and $\delta^{13}\text{C}_{\text{plant}}$ were 96.25 Mg ha^{-1} , 91.4 Mg ha^{-1} , -19.06‰ , -19.4‰ , and -28.0‰ , respectively. For Systems 1 and 2, the impact on C budgets and half-lives of ^{13}C isotopic discrimination (six hypothetical Rayleigh fractionation constants, -3.52 , -2.14 , -1.24 , 0 , 1.24 , and 2.40‰) during fresh organic matter and relic SOC mineralization was determined for a 13-yr period. For these calculations, it was assumed that 65% of the unharvested fresh biomass was mineralized.

In Model System 3, the potential impact on half-lives of landscape position and ^{13}C fractionation during mineralization was determined. Data previously reported by Clay et al. (2006) were used in this assessment. Soil organic C budgets were developed for SOC relic ϵ values of 0 and -2.52‰ . Clay et al. (2006) reported C budgets for this field when the Rayleigh fractionation constant was assumed to be 0‰. This scenario investigates the ramification of ^{13}C fractionation ($\epsilon = -2.52\text{‰}$) on these half-lives. As discussed in Clay et al. (2006), >600 soil samples from the 0- to 15-cm soil depth were collected from a 65-ha field located at $44^{\circ}10' \text{ N}$ and $96^{\circ}37' \text{ W}$. The samples were collected from a 30- by 30-m offset grid in May 1995 and between May and June in 2003. Each sample was a composite that contained 15 individual 1.7-cm-diameter cores collected every 11.4 cm along a transect. Soil samples were air dried (35°C), ground, sieved (2-mm sieve), and analyzed for total N, total C, $\delta^{15}\text{N}$, and ^{13}C discrimination (Δ) on a ratio mass spectrometer (Clay et al., 2003). Total C was corrected for inorganic C (Loeppert and Suarez, 1996). The soil samples from each year were aggregated to a common 40- by 40-m grid cell. The value of a grid cell was calculated as the average value of all the samples contained within a cell. A grid cell SOC value was the difference between inorganic (measured on the 1995 data set) and total C. As discussed in Clay et al. (2006), the grid cells were separated into the elevation zones <523.4, 523.4 to 527.3, 527.3 to 529.74, 529.74 to 532.2, and 532.2 to 534.30 m, which were approximately footslopes, toeslopes, backslopes, shoulder, and summit areas.

RESULTS AND DISCUSSION

Carbon-13 Discrimination during Relic Carbon Mineralization

At the Minnesota site, relic SOC values in the surface 30 cm were 90.8, 77.9, and 73.2 Mg ha^{-1} in 1980, 1993, and 2002, respectively. These values show that in areas where plant growth was prevented, SOC decreased 19.4% during 22 yr. Based on changes in total C during 22 yr, the relic SOC mineralization k value was 0.0098 and the half-life was 70.7 yr. The relic $\delta^{13}\text{C}$ SOC values at 0, 13, and 22 yr were -18.97 , -18.37 , and -18.25‰ . The Rayleigh fraction coefficients (ϵ) from 1980 to 1993 and from 1993 to 2002 were -3.91 and -1.92‰ , respectively. During the 22 yr of the experiment, ϵ averaged -3.45‰ . The value from 1993 to 2002 was similar to the long-term ϵ value of -1.71‰ reported for the Versailles experiment (Balesdent and Mariotti, 1996).

At the South Dakota research site, total soil C in the 0- to 15-cm depth averaged 26.8 g kg^{-1} (± 0.53) in 2000 and 24.0 g kg^{-1} (± 0.62) in 2005. Net loss of SOC was 2.8 g kg^{-1} (± 1.8) or 10.4% during 5 yr. Based on these values, the half-life was 31.4 yr. At this site, soil $\delta^{13}\text{C}$ averaged -17.19‰ (± 0.95) in 2000 and -16.64‰ (± 1.14) in 2005. The Rayleigh fractionation constant for this soil was -6.94‰ (± 4.74). Findings from South Dakota and Minnesota suggest that ^{13}C enrichment during relic SOC mineralization occurred and that the Rayleigh equation could be used to describe this enrichment process. Nedelhoffer and Fry (1988) had similar

Table 1. The potential influence of plant type on correlation coefficients between calculated half-life, relic C remaining in the soil after mineralization ($\text{SOC}_{\text{remaining}}$), the amount of relic C lost during mineralization (SOC_{lost}), and new C incorporated into SOC (PCR) and Rayleigh fractionation constants (ϵ) used to calculate ^{13}C isotopic discrimination during unharvested biomass (plant ϵ) and relic SOC ($\text{SOC } \epsilon$) mineralization.

Parameter	Plant type	Plant ϵ	SOC ϵ	Half-life
		<i>r</i>		
SOC ϵ	C ₄	0.00	1.00	
Half-life	C ₄	−0.41	−0.90	1.00
SOC _{remaining}	C ₄	−0.42	−0.76	0.91
SOC _{lost}	C ₄	0.42	0.76	−0.91
PCR	C ₄	0.42	0.76	−0.91
SOC ϵ	C ₃	0.00	1.00	
Half-life	C ₃	0.59	0.80	1.00
SOC _{remaining}	C ₃	0.56	0.58	0.80
SOC _{lost}	C ₃	−0.56	−0.58	−0.80
PCR	C ₃	−0.56	−0.58	−0.80

results and reported that the $\delta^{13}\text{C}$ value of bulk soil organic matter from forest mineral soils increased up to 0.5‰ during a 600-d period. Balesdent and Mariotti (1996) reported that, during a 60-yr period in an experiment initiated in 1928 at Versailles, France, relic SOC decreased 60% and $\delta^{13}\text{C}$ increased 1.6‰ at sites kept free of vegetation. Ueda et al. (2005) reported that $\delta^{13}\text{C}$ values increased with depth. The enrichment of relic C with depth and time has been attributed to respired CO_2 from soil microorganisms being depleted in ^{13}C (DeNiro and Epstein, 1978; Šantrůčková et al., 2000).

Carbon-13 Fractionation during Fresh Plant Biomass Mineralization

A 4-mo in-field incubation of corn and soybean residues was conducted to determine the fraction constant associated with fresh biomass. During the 4 mo, 31.5 (± 1.94) and 22.8 (± 0.44)% of 50 g of corn (C₄) and soybean (C₃) residues placed on the soil surface were mineralized, respectively. The mineralization of this material did not result in a measurable change in the $\delta^{13}\text{C}$ values of the corn and soybean residues. During the 4 mo, the corn residue C/N ratio increased from 42 to 57 and the C/N ratio in the soybean residue increased from 48 to 62. This increase was attributed to preferential mineralization of compounds with low C/N ratios. Others have also reported that ^{13}C enrichment during fresh plant biomass mineralization is insignificant.

Table 2. The influence of ^{13}C fractionation during SOC and unharvested biomass mineralization on the half-life, relic SOC residence time ($1/k$), relic C remaining in the soil ($\text{SOC}_{\text{remaining}}$), SOC lost (SOC_{lost}), and plant C incorporated into the soil ($\text{PCR}_{\text{incorporated}}$) after a simulated time of 13 yr.

Plant type	PCR ϵ	SOC ϵ	Half-life	Relic C residence time	SOC _{remaining}	SOC _{lost}	PCR _{incorporated}
	‰	‰	yr	yr/SOC	Mg ha ^{−1}		
C ₄	0	0	94	136	88	8.8	3.91
	0	−3.52	141	203	90	5.9	1.1
	−3.52	0	112	162	89	7.4	2.57
	−3.52	−3.52	149	217	91	5.6	0.79
C ₃	0	0	94	136	88	8.8	3.95
	0	−3.52	54	79	82	14.7	9.88
	−3.52	0	66	96	84	12.2	7.39
	−3.52	−3.52	22	32	64	32.5	27.7

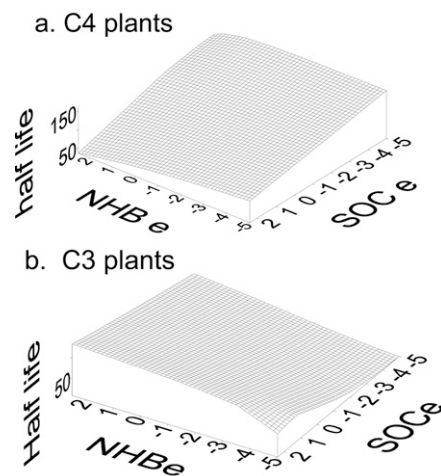


Fig. 1. The influence of the type of plant [(a) C₄ and (b) C₃] growing at a site and ^{13}C fractionation (ϵ) during the mineralization of fresh unharvested biomass returned to soil and soil organic C (SOC) on the calculated half-life of relic SOC.

Balesdent and Mariotti (1996) summarized the unpublished work of M. Linères (1996, INRA, Unite Agronomie, Laon, France), in which the $\delta^{13}\text{C}$ value of the initial corn biomass did not change after 85% of the biomass had been mineralized. Cleveland et al. (2004) reported that the $\delta^{13}\text{C}$ signatures of dissolved organic matter did not change during decomposition. Griebler et al. (2004) reported that ^{13}C fractionation of trichlorobenzene during mineralization was not observed under aerobic conditions but was observed under anaerobic conditions. Boutton (1996), in a review of isotopic ratios of SOC as indicators of change, stated that, “Direct measurements indicate that the $\delta^{13}\text{C}_{\text{PDB}}$ of plant tissue remains relatively constant during the early stages of decomposition (1–7 yr).” Fernandez and Cadisch (2003) reported that, with time, fractionation may even out, with microbes discriminating against ^{13}C (relative to the initial label) during early stages followed by a period of time when microbes discriminate against ^{12}C (relative to the initial label). The apparent lack of ^{13}C enrichment during the early stages of unharvested biomass mineralization may result from two independent processes cancelling each other out. The first factor is that many consumers of SOC tend to accumulate ^{13}C . The second factor is that materials that are resistant to microbial degradation (waxes and lignin) tend to be depleted in ^{13}C (Boutton, 1996; Huang et al., 1999; Conte et al., 2003).

Potential Impacts of Carbon-13 Discrimination Impacts on Half-Lives

Effects of C₃ and C₄ Residue

In a sensitivity analysis, the potential impacts of treating soil derived from C₃ and C₄ plants with C₄ residue and C₃ residue was assessed. For C₄ residue, the ϵ_{plant} and ϵ_{SOC} values were negatively correlated to half-lives and relic C remaining in the soil after mineralization (Table 1). For C₃ residue, the signs of the correlation coefficient were opposite those observed for C₄ residue.

Carbon-13 fractionation during SOC and unharvested biomass mineralization influenced the calculated relic C half-lives (Fig. 1, Table 2)

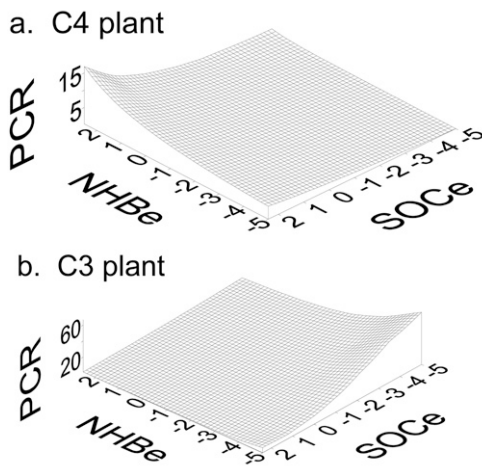


Fig. 2. The influence of plant type [(a) C_4 and (b) C_3] and ^{13}C fractionation during the mineralization of fresh unharvested biomass returned to soil and relic soil organic C (SOC) on the amount of fresh plant C retained (PCR) in the soil after mineralization.

and amount of plant carbon remaining in the soil after mineralization (PCR) (Fig. 2). When a C_4 plant was grown, half-lives of relic C ranged from 35 to 149 yr. Decreasing the ϵ_{SOC} and ϵ_{plant} values increased relic SOC half-lives and reduced the calculated amount of corn biomass incorporated into SOC (Table 2). For C_3 plants, different results were observed. Decreasing the ϵ_{SOC} decreased the calculated half-life, while decreasing ϵ_{PCR} increased relic C half-lives. The effect of ^{13}C fractionation on the amount of new C incorporated into the SOC mirrored the half-life results (Fig. 2).

Not considering ^{13}C fractionation in the relic SOC and unharvested biomass results in a cumulative error (Table 3). To further demonstrate the impact of this error, the ratio between $SOC_{retained}$ and SOC_{final} [$(\delta^{13}C_{soil\ final} - \delta^{13}C_{PCR}) / (\delta^{13}C_{SOC\ retained} - \delta^{13}C_{PCR})$] (derived from Eq. [6]) was determined for different assumptions, i.e., ^{13}C enrichment during mineralization occurred or did not occur. This ratio represents the relative proportion of SOC in the final sample after mineralization (Table 3). These calculations show that not considering ^{13}C fractionation, in either the relic soil or fresh biomass added to soil, results in cumulative errors. In systems where a C_4 plant was grown in soil derived from C_3 and C_4 plants, not considering ^{13}C fractionation results in underestimating the C derived from the relic C and overestimating the C from the fresh biomass. For C_3 plants, the reverse was true.

These calculations show that ^{13}C discrimination during mineralization had a consistent impact on calculated C budgets. Differences between the C_4 and C_3 plants were the direct result of ^{13}C discrimination on the relative difference between the $\delta^{13}C$ value of relic SOC after mineralization and the $\delta^{13}C$ value of the added residue. For C_4 plants, isotopic discrimination decreased this difference while for C_3 plants isotopic discrimination increased this difference.

Landscape Effects

To demonstrate the impact of ^{13}C discrimination on the interpretation of real data, SOC half-lives, using several ϵ values, were determined for data previously reported by Clay

Table 3. The hypothetical influence of ^{13}C isotopic discrimination on the calculated percentage of soil organic C (SOC) remaining in the soil after mineralization. The percentage of SOC retained relative to the amount of SOC at the end of the study (SOC_{final}) was derived from Eq. [6]. PCR initial is the of plant C returned to the soil.

Fractionation considered		δC _{SOC} value used					SOC retained
Soil	Fresh biomass	SOC _{final}	Initial SOC	SOC retained	PCR† initial	PCR retained	
		‰					%
no	no	−17	−19		−12		71.4
no	yes	−17	−19			−11	75.0
yes	no	−17		−18.5	−12		76.9
yes	yes	−17		−18.5		−11	80.0

† PCR = unharvested plant carbon returned to the soil.

et al. (2006). If ϵ_{SOC} was 0, then relic SOC half-lives ranged from approximately 50 yr in the tile-drained footslope area to 180 yr in the shoulder areas (Table 4). If ϵ was $-2.52‰$, however, then the calculated half-lives almost doubled. Associated with the increase in the half-life was a decrease in the contribution of unharvested biomass to SOC. In addition to influencing half-life calculations, ^{13}C discrimination will impact the mineralization rate constants calculated from these data, which in turn will impact any modeling effort designed to assess long-term changes. Clearly to accurately estimate SOC dynamics when using the ^{13}C natural abundance approach, an accurate estimate of ^{13}C enrichment during SOC mineralization is needed.

Landscape differences in SOC half-lives have clear implications for management. This is based on the assumption that relic SOC derived from C_3 and C_4 plants behave similarly. This assumption may not be valid. Henn and Chapela (2000) reported that ^{13}C discrimination during the mineralization of sucrose derived from C_3 plants increased with decreasing O_2 tension, and that fungi were ^{13}C enriched when cultured on C derived from C_3 sucrose and were not enriched when cultured on C_4 sucrose. Differences between C sources were attributed

Table 4. The influence of the Rayleigh fractionation coefficient (ϵ) and elevation zone on the initial amount of organic C contained in the soil at the beginning of the study ($SOC_{initial}$), the amount of relic C present in the soil after mineralization ($SOC_{retained}$), the amount of relic C lost during mineralization (SOC_L), the amount of plant C retained in the soil during the study (PCR), net change in C, and half-life.

Elevation zone	Relic SOC ϵ	$SOC_{initial}$	$SOC_{retained}$	SOC_L	PCR	Half-life
		Mg ha ⁻¹				yr
m	‰					
<523.4	0	52.7	44.0	8.73	6.23	49.8
523.4–527.3	0	52.2	44.4	7.75	7.42	56.1
527.3–529.74	0	49.1	45.4	3.76	2.78	113.1
529.74–532.2	0	49.1	46.7	2.39	2.95	181
532.2–534.2	0	48.6	43.4	5.24	6.32	78.9
<523.4	−2.52	52.7	47.6	5.06	2.56	89.1
523.4–527.3	−2.52	52.2	47.6	4.59	4.19	97.8
527.3–529.74	−2.52	49.1	47.2	1.87	0.97	232
529.74–532.2	−2.52	49.1	47.8	1.27	1.77	341
532.2–534.2	−2.52	48.6	45.8	2.80	3.91	151

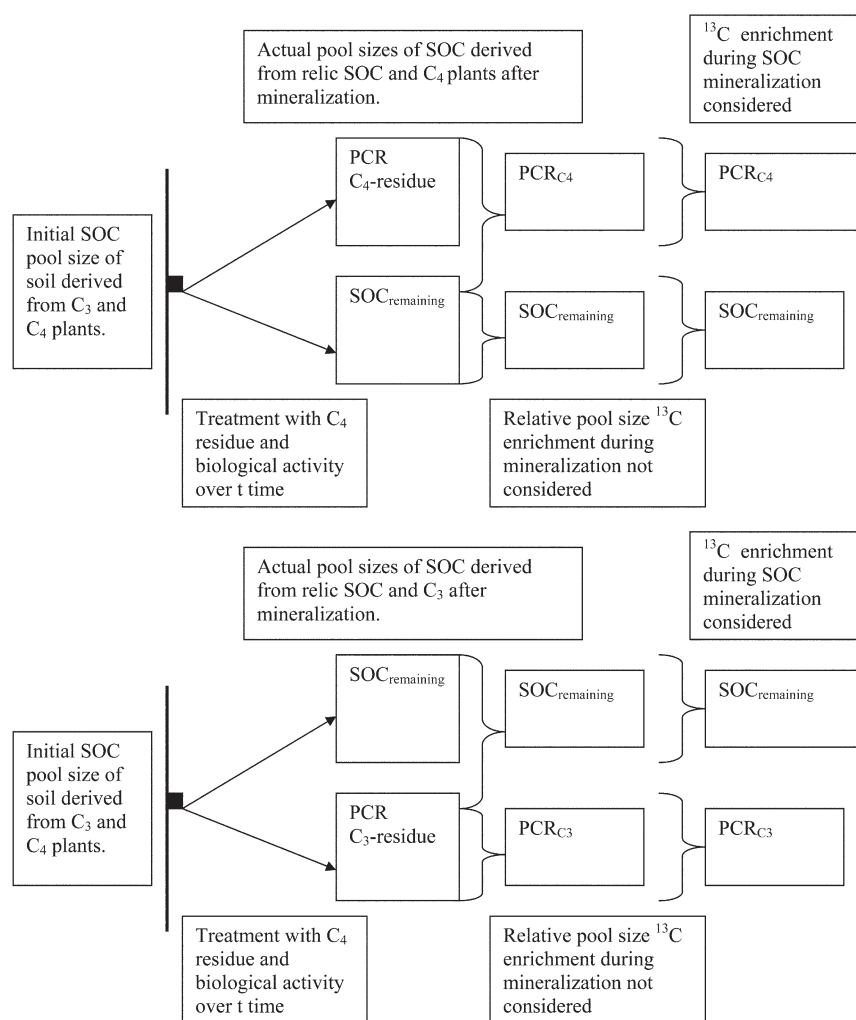


Fig. 3. Diagram showing the impact of plant type (C_3 and C_4 plants) and ^{13}C fractionation (Rayleigh fractionation coefficient $\epsilon = 0$ and $\epsilon < 0$) on the relative pool sizes of relic soil organic C (SOC) and unharvested biomass remaining in soil after mineralization. PCR = plant C retained.

to the ^{13}C atom not being randomly located in the sucrose molecule (Rossmann et al., 1991).

CONCLUSIONS

The measured SOC Rayleigh fractionation constants at sites located in South Dakota and Minnesota were negative, while the Rayleigh fractionation constant for NHC (ϵ_{PCR}) was not different from zero. The lack of ^{13}C enrichment during fresh residue mineralization can be attributed to two independent processes (^{13}C enrichment in consuming organisms and ^{13}C fractionation in plant biomass, i.e., lignin tends to be ^{13}C depleted relative to bulk soil) that tend to cancel each other out.

Others have reported isotopic enrichment during biological activity (Nedelhoffer and Fry, 1988; Balesdent and Mariotti, 1996; Boutton, 1996; Rochette et al., 1999; Clapp et al., 2000; Accoe et al., 2002; Fernandez and Cadisch, 2003; Griebler et al., 2004). The negative ϵ_{SOC} value for SOC mineralization was attributed to invertebrates and microbial biomass that feeds on SOC being ^{13}C enriched relative to the bulk soil (DeNiro and Epstein, 1978; Šantrůčková et al., 2000). The effect of ^{13}C discrimination on the SOC $\delta^{13}C$ value can be significant. For example, if ϵ is equal to -3.52 and 10% of the SOC is min-

eralized, then the $\delta^{13}C$ value of the relic C remaining after mineralization will increase 0.37‰ (Eq. [12]). Carbon-13 enrichment can have a profound impact on calculated half-lives and mineralization rate constants. These results were attributed to the impact of $\delta^{13}C$ discrimination on the calculated relative pool sizes of relic SOC and unharvested biomass C (Fig. 3).

Carbon-13 discrimination may partially explain lower mean residence time estimated using ^{13}C natural abundance than the ^{14}C dating approach (Paul et al., 2003). To account for ^{13}C discrimination during mineralization, experiments should contain controls where ^{13}C fractionation is measured. The effects of ^{13}C enrichment during relic SOC and unharvested biomass mineralization were cumulative and did not cancel each other out. Findings from this study suggest that experiments that do not account for ^{13}C enrichment will overestimate the contribution of C_4 biomass to SOC, underestimate the contribution of C_3 biomass to SOC, and underestimate SOC stability.

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